

Amendments to the Claims:

This listing of claims will replace all prior versions in the present application:

Listing of Claims:

1. (withdrawn) An isolated human nuclear receptor that binds to a cytochrome P-450 monooxygenase promoter, or a DNA binding or ligand binding domain thereof.

2. (withdrawn) The receptor according to claim 1 wherein the promoter is a cytochrome P-450 monooxygenase 3A4 (CYP3A4) promoter.

3. (withdrawn) The receptor according to claim 2 wherein said receptor is hPXR.

4. (withdrawn) An isolated human nuclear receptor having the amino acid sequence given Figure 1, or a fragment thereof, of at least 30 consecutive amino acids.

5. (withdrawn) A fusion protein comprising a DNA binding or ligand binding domain of hPXR and a non-hPXR-derived sequence.

6. (withdrawn) An isolated nucleic acid comprising a sequence encoding the receptor of claim 1 or 4 or the fusion protein of claim 5.

7. (withdrawn) A construct comprising the nucleic acid of claim 6 and a vector.

8. (withdrawn) A host cell comprising the construct of claim 7.

9. (withdrawn) A method of making the receptor of claim 3, or fragment thereof, comprising:

culturing a host cell containing an expression construct comprising a sequence encoding said receptor, or fragment thereof, operably linked to a promoter, under conditions such that said receptor, or fragment thereof, is produced, and

isolating said receptor, or fragment thereof.

10. (Amended) A method of screening a test compound for its ability to induce cytochrome P-450 3A4 (CYP3A4) gene expression comprising:

(i) contacting said test compound with a protein comprised of a ligand binding domain of human pregnane X receptor (hPXR) having the amino acid sequence 141-434 of SEQ ID NO:14, wherein the protein shares at least 96% amino acid sequence identity with the ligand binding domain of SEQ ID NO:14 and retains the sequence's ligand-binding function,

(ii) determining whether said test compound selectively binds to the ligand binding domain of said protein; and

(iii) determining whether a test compound that selectively binds to the ligand binding domain of said protein induces receptor binding to a response element in the CYP3A4 gene promoter and CYP3A4 enzyme expression of a cytochrome P-450 3A4 monooxygenase enzyme.

11. (withdrawn) A method of screening a test compound for its ability to activate or inhibit hPXR comprising:

(i) preparing an expression vector comprising a sequence encoding a DNA binding domain and a hPXR ligand binding domain;

(ii) preparing a reporter construct comprising a DNA binding site recognized by said DNA binding domain operably linked to a reporter gene;

(iii) introducing said expression vector and said reporter construct into compatible host cells;

(iv) incubating said cells resulting from step (iii) with said test compound, and

(v) determining the level of expression of said reporter gene, wherein enhancement of expression of said reporter gene in the presence of said test compound indicates that said test compound can activate hPXR, and

wherein inhibition of expression of said reporter gene in the presence of said test compound indicates that said test compound can inhibit hPXR.

12. (withdrawn) A compound that induces CYP3A4 identified by the method of claim 10.

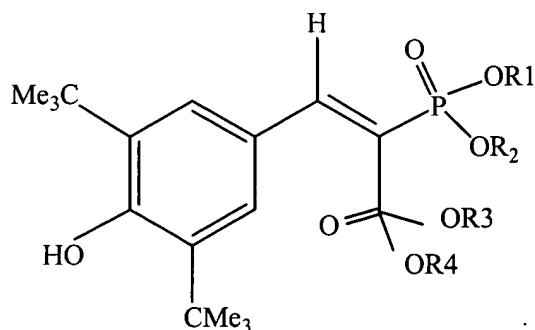
13. (withdrawn) A compound that activates hPXR identified by the method of claim 11.

14. (withdrawn) A method of modulating function of a cell mediated by PXR comprising contacting said cell with a compound identified using the method of claim 11 that activates PXR under conditions such that said activation is effected and said function is thereby modulated.

15. (withdrawn) A method of modulating function of a cell mediated by PXR comprising contacting said cell with a compound identified using the method

of claim 11 that inhibits PXR under conditions such that said inhibition is effected and said function is thereby modulated.

16. (withdrawn) The method according to claim 14 or 15 wherein said compound is of formula I:



wherein R1, R2, R3 and R4 are, independently, C₁-C₆ alkyl, linear or branched.

17. (withdrawn) The method according to claim 14 or 15 wherein said cell is a cultured cell.

18. (withdrawn) The method according to claim 14 or 15 wherein said cell is present in a tissue.

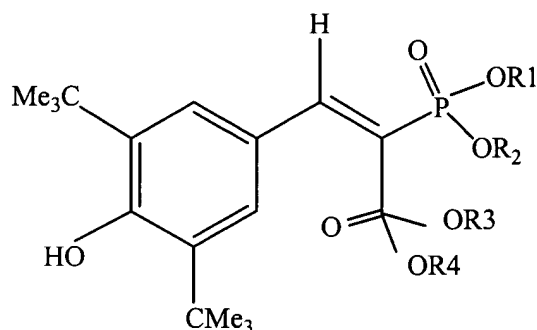
19. (withdrawn) The method according to claim 14 or 15 wherein said cell is present in an animal.

20. (withdrawn) A method for associating a particular disease or condition with modulation of PXR comprising

contacting a compound that binds to PXR specifically with PXR present in a cell under conditions such that said binding is effected and functional activity of said cell mediated by PXR is thereby modulated,

detecting said modulation of said functional activity and associating said modulation of said functional activity with a disease or condition and thereby associating said disease or condition with modulation of PXR.

21. (withdrawn) The method according to claim 20 wherein said compound is of formula I:



wherein R1, R2, R3 and R4 are, independently, C₁-C₆ alkyl, linear or branched.

22. (withdrawn) The method according to claim 21 wherein said compound is GW-485801.

23. (withdrawn) A method of preventing or treating a disease or condition that has been associated with modulation PXR by the method of claim 20, comprising administering to a patient in need thereof a therapeutically effective amount of an agent that modulates the activity of PXR so that said prevention or treatment is effected.

24. (withdrawn) The method according to claim 23 wherein said agent is GW-485801.

25. (Amended) The method according to claim 10 which, wherein the method is an in vitro assay.

26. (withdrawn) The method according to claim 10 which is an in vivo assay.

27. (Amended) The method according to claim 10 wherein ~~said protein has an amino acid sequence including amino acids 141 to 434 of SEQ ID NO: 14~~: the protein shares at least 97% amino acid sequence identity with the ligand binding domain of SEQ ID NO: 14 and retains the sequence's ligand-binding function.

28. (Previously presented) The method according to claim 10 wherein said protein has an amino acid sequence including amino acids 130 to 434 of SEQ ID NO: 14.

29. (Amended) The method according to claim 10 wherein said protein ~~has an amino acid sequence including SEQ ID NO: 14~~ shares at least 98% amino acid sequence identity with the ligand binding domain of SEQ ID NO: 14 and retains the sequence's ligand-binding function.

30. (Previously presented) The method according to claim 10 wherein said protein bears a detectable label.

31. canceled.

32. canceled.

33. canceled.

34. (Amended) The method according to claim 10 wherein ~~said protein is a chimeric receptor~~ the ligand-binding domain of an hPXR polypeptide is fused to a DNA binding domain of a non-hPXR polypeptide.

35. canceled.

36. (withdrawn) The method according to claim 25 wherein said protein is bound to a solid support.

37. (Previously presented) The method according to claim 25 wherein binding is determined by separating test compound bound to protein from free test compound and free protein.

38. (Amended) The method according to claim 10 wherein binding is determined by a scintillation proximity assay.

39. (Previously presented) The method according to claim 10 wherein binding is determined by competitive binding assay.

40. (withdrawn) A method of selecting a drug compound which does not induce cytochrome P450 3A4 (CYP3A4) gene expression comprising:

(i) determining whether a drug compound induces CYP3A4 gene expression in the presence of a protein comprised of a ligand binding domain of human pregnane X receptor (hPXR) having the amino acid sequence of SEQ ID NO: 14, and

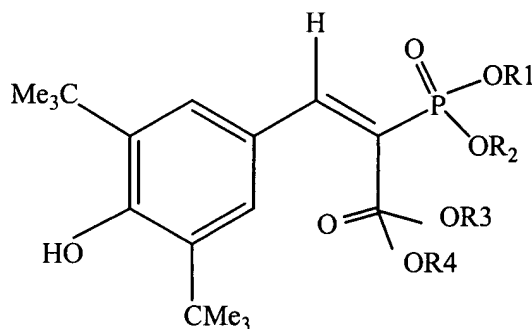
(ii) selecting a drug compound which does not induce CYP3A4 gene expression.

41. (Amended) A method of screening a test compound for its ability to bind to a protein comprising human pregnane X receptor ligand binding domain, thereby indicating an increased likelihood that the test compound alters in vivo expression of the a cytochrome P-450 3A4 (CYP3A4) monooxygenase enzyme comprising:

(i) contacting said test compound with a protein comprised of a ligand binding domain of human pregnane X receptor (hPXR), said ligand binding domain including the amino acid sequence of amino acids 141-434 of SEQ ID NO:14, wherein the protein comprises a domain sharing an amino acid sequence at least 96% identical to the ligand binding domain of SEQ ID NO: 14, and

(ii) determining whether said test compound selectively binds to the ligand binding domain of said protein.

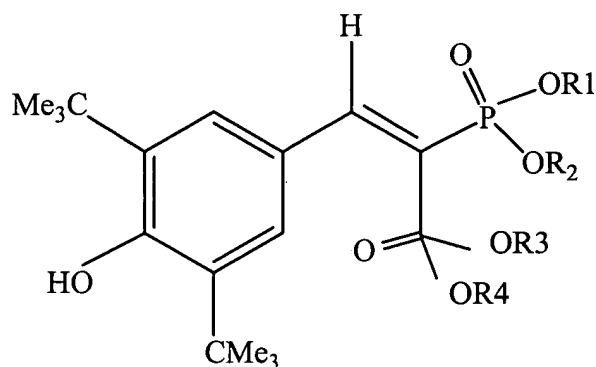
42. (new) The method according to claim 39, wherein a test compound of formula 1 is detectably labeled



and each of R₁, R₂, R₃ and R₄ is, independently, C₁-C₆ alkyl (linear or branched).

43. (new) A method for identifying a compound as an hPXR agonist, the method comprising:

providing a polypeptide comprising the ligand-binding domain of an hPXR, wherein the ligand-binding domain comprises amino acids 130-434 of SEQ ID NO: 14, wherein the polypeptide selectively binds a detectably labeled compound of formula 1



and each of R1, R2, R3 and R4 is, independently, C1-C6 alkyl (linear or branched);

contacting the polypeptide with a test compound;

determining whether the binding of the polypeptide to the detectably labeled compound of formula 1 is altered in the presence of the test compound, a decrease in the binding being an indication that the test compound is a competitive inhibitor of the detectably labeled compound of formula 1; and

determining whether expression of a CYP3A4 gene product, following receptor binding to a response element in the CYP3A4 gene promoter, is altered in a cell in the presence of the test compound, wherein an increase in the expression is an indication that the test compound is useful as an hPXR agonist in screening assays.

44. (new) The method according to claim 42 or 43, wherein the detectably labeled compound of formula I is GW-485801.

45. (new) The method according to claim 43, wherein the cytochrome P450 3A4 gene product is a cytochrome P-450 3A4 monooxygenase enzyme.

46. (new) The method of claim 10, wherein the ligand-binding domain is a fragment of SEQ ID NO: 14 at least 75 consecutive amino acid residues in length.

47. (new) The method of claim 10, wherein the ligand-binding domain is a fragment of SEQ ID NO: 14 at least 50 consecutive amino acid residues in length.

48. (new) The method of claim 10, wherein the ligand-binding domain is a fragment of SEQ ID NO: 14 at least 30 consecutive amino acid residues in length.